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Comparison of Outcomes for Pediatric Patients With Acute Myeloid Leukemia in Remission and Undergoing Allogeneic Hematopoietic Cell Transplantation With Myeloablative Conditioning Regimens Based on Either Intravenous Busulfan or Total Body Irradiation: A Report From the Japanese Society for Hematopoietic Cell Transplantation

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A B S T R A C T

Pediatric patients with acute myeloid leukemia (AML) mainly receive myeloablative conditioning regimens based on busulfan (BU) or total body irradiation (TBI) before allogeneic hematopoietic cell transplantation (allo-HCT); however, the optimal conditioning regimen remains unclear. To identify which of these regimens is better for pediatric patients, we performed a retrospective analysis of nationwide registration data collected in Japan between 2006 and 2011 to assess the outcomes of patients receiving these regimens before a first allo-HCT. Myeloablative conditioning regimens based on i.v. BU (i.v. BU-MAC) (n = 69) or TBI (TBI-MAC) (n = 151) were compared in pediatric AML patients in first or second complete remission (CR1/CR2). The incidences of sinusoid obstruction syndrome, acute and chronic graft-versus-host disease, and early non-relapse mortality (NRM) before day 100 were similar for both conditioning groups; however, the incidence of bacterial infection during the acute period was higher in the TBI-MAC group ($P = .008$). Both groups showed a similar incidence of NRM, and there was no significant difference in the incidence of relapse between the groups. Univariate and multivariate analyses revealed no significant differences in the 2-year relapse-free survival rates for the i.v. BU-MAC and TBI-MAC groups in the CR1/CR2 setting (71% versus 67%, $P = .36$;

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hazard ratio, .73; 95% CI, .43 to 1.24, respectively). TBI-MAC was no better than i.v. BU-MAC for pediatric AML patients in remission. Although this retrospective registry-based analysis has several limitations, i.v. BU-MAC warrants further evaluation in a prospective trial.

INTRODUCTION

Intensive combination chemotherapy results in a 52% to 75% probability of survival for childhood and adolescent patients with acute myeloid leukemia (AML); however, more than 30% of patients relapse [1-4]. Although allogeneic hematopoietic cell transplantation (allo-HCT) is the most promising therapy for intractable disease (eg, cytogenetically unfavorable disease and relapsed disease), the conditioning regimen (as well as effective control of graft-versus-leukemia effects) plays an important role in reducing the incidence of relapse after transplantation [5]. Although pediatric AML patients often receive myeloablative conditioning (MAC) regimens based on total body irradiation (TBI) or busulfan (BU), no optimal regimen has been devised.

Myeloablative TBI conditioning regimens are associated with late complications, which manifest as growth retardation, neurocognitive effects, cataracts, hypothyroidism, gonadal dysfunction, infertility, and a significantly increased risk of a second malignancy [6,7]. Myeloablative BU conditioning regimens can also result in some of these late complications, although the incidence of growth retardation, neurocognitive effects, cataracts, thyroid dysfunction, and second malignancies may be lower [7-10]. Recent studies show treatment of adult AML patients with i.v. BU results in better survival than treatment with TBI [11,12]; however, few reports have examined these regimens in children. Sisler et al. [13] showed conditioning regimens that include TBI have no additional benefit of over those that include BU in pediatric patients beyond first complete remission (CR1). A report by de Berranger et al. [14] demonstrated that disease-free survival was significantly better after BU and cyclophosphamide (CY) than after TBI and CY. However, it should be noted that the patients in these studies received both i.v. and oral BU as a MAC regimen. Also, approximately half of the patients in the latter study received HCT before 2000. On the other hand, another study showed that i.v. BU failed to provide a significant survival advantage in children with acute leukemia when compared with oral BU [15].

The efficacy and adverse events associated with i.v. BU-MAC regimens are unclear, particularly when used to treat pediatric AML patients. Therefore, the present study aimed to compare the outcomes for pediatric AML patients after i.v. BU-MAC or TBI-MAC.

METHODS

Patients and Transplantation

Pediatric patients (aged <18 years) with de novo AML (excluding AML-M3) who underwent a first allo-HCT after either i.v. BU- or TBI-based MAC in CR1 or second CR (CR2) between January 2007 and December 2012 were recruited for the study. Patients were prospectively enrolled in the Japanese Data Center for Hematopoietic Cell Transplantation. Patients with Down's syndrome, Fanconi anemia, or neurofibromatosis type 1 and those who received a graft after ex vivo T cell depletion or $CD34^+$ selection or a graft from an HLA-haploidentical donor were excluded. Patients who received combination regimens comprising TBI and BU were also excluded. Finally, data from consecutive patients who received HCT after myeloablative TBI combined with cytotoxic drugs (TBI-MAC) or myeloablative i.v. BU combined with cytotoxic drugs (BU-MAC) were examined. MAC regimens were defined as regimens that included either fractionated TBI >8 Gy or i.v. BU >6.4 mg/kg [16].

Unfavorable cytogenetics/genetics were defined as either 7-/7q-, 5q-, complex karyotype, t(6;11), t(6,9), t(16;21), t(9:22), or as fms-like tyrosine kinase receptor 3 internal tandem duplication. Favorable genetics were defined as either t(8;21) or inv(16). Intermediate genetics were defined as neither unfavorable nor favorable [17,18]. Graft-versus-host disease (GVHD) was graded according to previously published and accepted criteria [19]. Nonrelapse mortality (NRM) was defined as death during continuous remission, and relapse-free survival (RFS) was defined as survival without any relapse of the underlying hematological malignancy or death from any cause.

Statistical Analysis

Pair-wise comparisons of patient, disease, and transplant characteristics (covariates) were performed using Fisher's exact test (for categorical variables). Variables considered in the analysis included year of transplantation (2007 to 2009 versus 2010 to 2012), gender, age at the time of transplantation (<10 versus ≥10), FAB classification (M0, M1, and M2 versus M4 and M5 versus M6 and M7), disease status (CR1 versus CR2), cytogenetics/genetics risk category (favorable versus others), extramedullary/central nervous system involvement (negative versus positive), donor status (matched sibling donor versus others), graft source (bone marrow versus peripheral blood stem cells versus cord blood), donor–recipient HLA-A, -B, and -DR antigen matching (match versus mismatch to GVH direction), donor–recipient ABO group matching (major matching versus major mismatching), donor–recipient gender matching (female-to-male versus other combinations), donor–recipient cytomegalovirus status (negative-to-negative versus other combinations), GVHD prophylaxis (cyclosporine-based prophylaxis versus others), performance status (<2 versus ≥2), comorbidity index (<2 versus ≥2), and conditioning regimen (i.v. BU-MAC versus TBI-MAC). Survival was estimated using the Kaplan-Meier method and log-rank tests, whereas the cumulative incidence between groups was analyzed using Gray's test.

Risk factors associated with relapse, NRM, RFS, and conditioning group were assessed using multivariate Cox and Fine-Gray proportional-hazard models. Parameters with $P < .2$ on univariate analysis were included in the model. NRM was the competing event for relapse, and relapse was the competing event for NRM. Any incidence of death or relapse was the competing event for GVHD onset.

Statistical analyses were performed using STATA 12 (Stata Corp., TX) and EZR data analysis programs [20]. Statistical significance was set at $P < .05$. The present study had 71% power for detecting a 15% difference in the 2-year survival rate between the i.v. BU-MAC and TBI-MAC groups with an error (2-sided) of .05 [20]. The study was approved by the institutional review boards of Matsushita Memorial Hospital and by the Japanese Society for Hematopoietic Cell Transplantation committee.

RESULTS

Patient Characteristics

Data from 220 patients who received either i.v. BU-MAC (n = 69) or TBI-MAC (n = 151) were analyzed in detail. Intravenous BU-MAC comprised BU plus CY or BU plus melphalan (MEL) either with or without another cytotoxic drug (n = 13 and n = 52, respectively) or other miscellaneous combinations, including BU (n = 4), whereas TBI-MAC comprised TBI plus CY or TBI plus MEL either with or without another cytotoxic drug (n = 107 and n = 42, respectively) or other miscellaneous combinations, including TBI (n = 2). Preliminary analyses revealed that RFS after TBI and CY ± another cytotoxic drug was similar to that after TBI and MEL ± another cytotoxic drug (2-year-RFS: 65% versus 71%, respectively; $P = .51$) and that RFS after i.v. BU and CY ± another cytotoxic drug was similar to that after i.v. BU and MEL ± another cytotoxic drug (2-year-RFS: 71% versus 71%, respectively; $P = .87$). We therefore compared the outcomes after i.v. BU-MAC with those after TBI-MAC.

Table 1 summarizes patient characteristics, comorbidities, and transplant procedures for each conditioning group.

Table 1
Demographic and Clinical Characteristics According to Conditioning Regimen

	i.v. BU-MAC n (%)	TBI-MAC n (%)	Total n (%)	*P*
Patient characteristics	69 (31)	151 (69)	220 (100)	
Year of transplant				.56
2007-2009	37 (54)	74 (49)	111 (50)	
2010-2012	32 (46)	77 (51)	109 (50)	
Gender				.25
Female	31 (45)	82 (54)	113 (51)	
Male	38 (55)	69 (46)	107 (49)	
Age at the time of transplant				<.001
≤4 yr	44 (64)	23 (15)	67 (30)	
5-9 yr	10 (15)	38 (25)	48 (22)	
≥10 yr	15 (22)	90 (60)	105 (48)	
FAB classification				
M0	4 (6)	15 (10)	19 (9)	
M1	7 (10)	29 (19)	36 (16)	
M2	10 (14)	58 (38)	68 (31)	
M4	8 (12)	18 (12)	26 (12)	
M5	12 (17)	16 (11)	28 (13)	
M6	4 (6)	6 (4)	10 (5)	
M7	19 (28)	5 (3)	24 (11)	
Others*/missing data	5 (7)	4 (3)	9 (4)	
M0–M2 vs. M4, M5 vs. M6, M7				<.001
M0–M2	21 (30)	102 (68)	123 (56)	
M4, M5	20 (29)	34 (23)	54 (24)	
M6, M7	23 (33)	11 (7)	34 (15)	
Others*/missing data	5 (7)	4 (3)	9 (4)	
Disease status at the time of transplant				.01
CR1	55 (80)	94 (62)	149 (68)	
CR2	14 (20)	57 (38)	71 (32)	
Cytogenetics/genetics				.055
Favorable	6 (9)	33 (22)	39 (18)	
Intermediate	46 (67)	92 (61)	138 (63)	
Unfavorable	16 (23)	25 (17)	41 (19)	
UA/missing data	1 (1)	1 (1)	2 (1)	
Extramedullary involvement/CNS involvement				.18/.002
Negative	56/45 (89/65)	140/128 (93/85)	196/173 (89/78)	
Positive CNS/total	13/24 (19/35)	11/23 (7/15)	24/47 (11/22)	
Donor				.49
Matched sibling	17 (25)	31 (21)	48 (22)	
Others	52 (75)	120 (79)	172 (78)	
Graft				.95
Bone marrow	40 (58)	84 (56)	124 (56)	
Peripheral blood stem cell	6 (9)	15 (10)	21 (10)	
Cord blood	23 (33)	52 (34)	75 (34)	
HLA				
Match	46 (67)	81 (54)	127 (58)	.08
Mismatch	23 (33)	70 (46)	93 (42)	
ABO group				.64
Major match	22 (32)	43 (28)	65 (3)	
Major mismatch	47 (68)	108 (72)	155 (70)	
Gender				.03
Female to male	20 (29)	24 (16)	44 (20)	
Others	37 (54)	107 (71)	144 (65)	
Missing data	12 (17)	20 (13)	32 (15)	
Cytomegalovirus status				.76
Negative to negative	9 (13)	18 (12)	27 (12)	
Others	53 (77)	112 (74)	165 (75)	
Missing data	7 (10)	21 (14)	28 (13)	
GVHD prophylaxis				.08
Cyclosporine-based	21 (30)	48 (32)	69 (31)	
Tacrolimus-based	44 (64)	102 (68)	146 (67)	
Others	4 (6)	1 (1)	5 (2)	
Performance status at the time of transplant				.03
<2	63 (91)	148 (98)	211 (96)	
≥2	6 (9)	3 (2)	9 (4)	
HCT-CI points				.79
<2	59 (86)	132 (87)	191 (86)	
≥2	2 (3)	6 (4)	8 (4)	
UA/missing data	8 (12)	13 (9)	21 (10)	
Median observation period, mo				
	31 (4-78)	27 (.3-80)	28 (.3-80)	

UA indicates unassessable; CNS, central nervous system; HCT-CI, hematopoietic cell transplantation-specific comorbidity index.
* AML with multilineage dysplasia or acute undifferentiated leukemia.

Thirty-one percent (n = 69) and 69% (n = 151) of patients received HCT after i.v. BU-MAC and TBI-MAC, respectively. A total of 10 Gy TBI in 4 or 5 fractions (1 recipient each); 12 Gy TBI in 4 (9 recipients), 5 (6 recipients), or 6 fractions (133 recipients); and 13.2 Gy TB1 in 6 fractions (1 recipient) was administered. However, we have no information about whether BU was targeted to a certain level. Only 2 patients in each conditioning group received antithymocyte globulin as part of the conditioning regimen. The median age at the time of transplantation was 9 years (range, 0 to 17 years). Of the 218 patients (99%) with assessable cytogenetics and/or genetics, 18%, 63%, and 19% fell into the favorable, intermediate, and unfavorable categories, respectively. Four patients who had favorable cytogenetics without induction delay underwent HCT in CR1, despite the absence of any formal indications. The median interval from diagnosis to transplantation was 5.5 months for those in CR1 and 19 months for those in CR2. The median follow-up time was 28 months (range, .3 to 80). There were significant differences between groups in terms of age at the time of transplantation ($P < .001$), FAB classification ($P < .001$), disease status ($P = .01$), extramedullary involvement ($P = .002$), recipient and donor gender combination ($P = .03$), and performance status ($P = .03$).

There was no difference in the actuarial incidence of engraftment, sinusoidal obstruction syndrome, and cumulative incidence of NRM up until day 100 between the 2 groups. The graft source for 5 of 6 engraftment failure cases was cord blood. The group conditioned with i.v. BU-MAC experienced fewer bacterial infectious than the group conditioned with TBI-MAC (30% versus 50%, respectively; $P = .008$). The cumulative incidence of grades II to IV acute GVHD was 32% (95% confidence interval [CI], 21% to 43%) and 36% (95% CI, 29% to 44%) ($P = .46$) and of extensive chronic GVHD was 12% (95% CI, 6% to 21%) and 12% (95% CI, 7% to 17%) ($P = .91$) in the i.v. BU-MAC and TBI-MAC groups, respectively (Table 2, Figure 1A,B).

The 2-year cumulative incidences of relapse and NRM were 24% (95% CI, 18% to 30%) and 8% (5% to 12%), respectively, and the 2-year RFS rate was 68% (95% CI, 61% to 74%): 67% (95% CI, 59% to 74%) in CR1 and 69% (95% CI, 5% to 79%) in CR2. The main cause of death was relapse (57% in the BU-MAC group and 51% in the TBI-MAC group). Both groups were balanced with respect to cause of death (Table 3; $P = .75$).

Incidence of Relapse and NRM after Each Conditioning Regimen

The 2-year cumulative incidence of relapse in the i.v. BU-MAC group was 24% (95% CI, 15% to 35%) and in the TBI-MAC group was 24% (95% CI, 17% to 31%) ($P = .95$); the cumulative incidences of NRM were 5% (95% CI, 1% to 13%) and 10% (95% CI, 6% to 15%) ($P = .12$), respectively (Figure 2A,B). The rate of relapse differed significantly according to cytogenetics/genetics in that the 2-year cumulative incidence of relapse was 11% (95% CI, 3% to 23%) in the favorable cytogenetics/genetics group versus 27% (95% CI, 20% to 34%) in the nonfavorable cytogenetics/genetics group ($P = .049$). NRM differed significantly according to HLA matching in that the 2-year cumulative incidence of NMR was 3% (95% CI, 1% to 8%) in the HLA-matched group versus 15% (95% CI, 9% to 24%) in the HLA-mismatched group ($P = .003$).

Multivariate analysis revealed no significant association between the rates of relapse or NRM after i.v. BU-MAC or TBI-MAC (relapse: hazard ratio [HR], .85; 95% CI, .47 to 1.54; $P = .59$; NRM: HR, .70; 95% CI, .18 to 2.78; $P = .61$, respectively). However, NRM rates were significantly higher in the HLA-mismatched group (HR, 3.87; 95% CI, 1.15 to 13.1; $P = .03$).

Table 2
Transplantation Outcomes

	i.v. BU-MAC (n = 69)	TBI-MAC (n = 151)	*P*
Engraftment			.44
No	1	4	
Yes	99	96	
Bacterial infection			.008
No	70	50	
Yes	30	50	
Sinusoidal obstruction syndrome			.55
No	30	23	
Yes	3	4	
UA/missing data	67	73	
Day 100 NRM			.06
No	100	95	
Yes	0	5	
Acute GVHD (day 100)			
Grades II-IV	32 (21-43)	36 (29-44)	.46
Grades III-IV	17 (10-27)	14 (9-20)	.53
Chronic GVHD (2 year)			
All	33 (22-45)	23 (17-30)	.12
Extensive type	12 (6-21)	12 (7-17)	.91
Cumulative incidence of relapse* (2 year)	24 (15-35)	24 (17-31)	.95
Cumulative incidence of NRM† (2 year)	5 (1-13)	10 (6-15)	.12
RFS‡ (2 year)	71 (58-80)	67 (58-74)	.36

Values are percents with 95% CIs in parentheses. Factors with P < .2 in the univariate model are denoted by symbols.

* ABO group major match (match vs. mismatch, $P = .08$), Cytogenetics/genetics (favorable vs. intermediate or unfavorable, $P = .04$), and performance status (<2 vs. ≥2, $P = .09$).

† Age (<10 yr vs. ≥10 yr, $P = .17$), ABO group major match (match vs. mismatch, $P = .14$), extramedullary involvement (negative vs. positive, $P = .08$), HLA match (match vs. mismatch, $P = .003$), GVHD prophylaxis (cyclosporine-based vs. other prophylaxis, $P = .17$), donor (matched sibling donor vs. others, $P = .08$), and year of transplant (2007-2009 vs. 2010-2012, $P = .09$).

‡ Cytogenetics/molecular marker (favorable vs. intermediate or unfavorable, $P = .10$) and HLA match (match vs. mismatch, $P = .16$).

RFS after Each Conditioning Regimen

There was no difference in 2-year RFS in the setting of CR1/CR2 (71% [95% CI, 58% to 80%] versus 67% [95% CI, 58% to 74%] for the TBI-MAC and TBI-MAC groups, respectively; $P = .36$) (Figure 2C). The overall survival rates in the i.v. BU-MAC and the TBI-MAC groups were 80% (95% CI, 68% to 88%) and 73% (95% CI, 65% to 80%), respectively, ($P = .12$).

Multivariate analysis revealed no significant differences in the RFS rates for the i.v. BU-MAC and TBI-MAC groups (HR, .73; 95% CI, .43 to 1.24; $P = .24$). The RFS rates for the intermediate and unfavorable cytogenetics/genetics groups (HR, 2.34; 95% CI, 1.06 to 5.15; $P = .03$) were significantly lower than those for the favorable group.

RFS According to Other Factors

There was no difference in RFS between grafts from matched sibling donors and those from unrelated donors when transplanted after BU-MAC (2-year RFS: 58% versus 75%, respectively; $P = .15$) and when transplanted after TBI-MAC (2-year RFS: 74% versus 65%, respectively; $P = .37$). Although the number of patients was small, the RFS for patients with central nervous system disease or advanced disease (>CR1) was similar after TBI-MAC and BU-MAC (2-year RFS: 64% versus 77%; $P = .54$, and 68% versus 71%; $P = .80$, respectively).

DISCUSSION

Prospective [21], retrospective [22,23], and meta-analysis [24] studies provide no clear answer regarding the

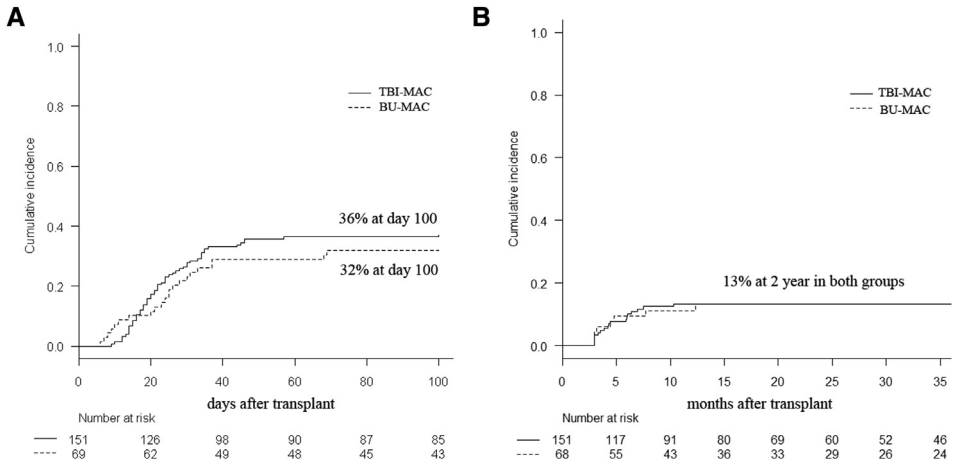

Figure 1. (A) The cumulative incidence of grades II to IV acute GVHD was 32% (95% CI, 21% to 43%) in the i.v. BU-MAC group (n = 69; dotted line) and 36% (95% CI, 29% to 44%) ($P = .46$) in the TBI-MAC group (n = 151; solid line). (B) The cumulative incidence of extensive chronic GVHD was 12% (95% CI, 6% to 21%) in the i.v. BU-MAC group (dotted line) and 12% (95% CI, 7% to 17%; $P = .91$) in the TBI-MAC group (solid line).

superiority of TBI-MAC over BU-MAC; however, such studies suggest that adult AML patients conditioned with TBI-MAC experience a lower incidence of relapse [21-24]. A meta-analysis of 18 trials (3172 leukemia patients) indicated that TBI-MAC regimens lead to lower rates of relapse in patients with AML (713 patients in 3 trials); higher rates of NRM in patients with acute lymphoblastic leukemia, AML, or chronic myeloid leukemia (2586 patients in 8 trials); and higher rates of disease-free survival in patients with AML (1289 patients in 4 trials) [25]. However, all these studies mainly recruited patients who were conditioned with oral BU. A recent retrospective analysis indicated that i.v. BU-MAC yielded similar RFS and overall survival rates to TBI-MAC when used to condition adult AML patients (over 18 years old); however, although the rates of NRM were similar, the rates of relapse were higher [26]. Another retrospective analysis from the Center for International Blood and Marrow Transplant Research showed that for patients with AML in CR1, i.v. BU and CY conditioned groups had better RFS and overall survival than TBI and CY conditioned groups, with similar rates of relapse and a better NRM [12]. Furthermore, Bredeson et al. [11] conducted a prospective nonrandomized trial involving patients with AML, myelodysplastic syndrome, or chronic myelogenous leukemia in remission (all patients were ≤60 years old) and showed that i.v. BU-MAC resulted in superior overall survival (but not RFS) without an increased risk of relapse or NRM.

However, the utility of i.v. BU-MAC for pediatric AML patients remains unclear. In the field of pediatric AML, Sisler et al. [13] showed that TBI for pediatric AML patients beyond CR1 (n = 151) had no advantages over oral or i.v. administration of BU. Recently, a French group compared BU and TBI for pediatric AML cases in CR1 (n = 226) and found survival was significantly better for those receiving oral or i.v. BU (16 mg/kg for oral BU or i.v. BU adapted to recipient body weight) and CY (200 mg/kg) than for those receiving TBI (fractionated, 12 Gy) and CY (120 mg/kg), with a more favorable NRM [14]. Here, we expected to find superior (or at least comparable) outcomes for the i.v. BU-MAC group; however, we found that the incidences of relapse, NRM, and RFS after i.v. BU-MAC were similar to those after TBI-MAC in the setting of CR1/CR2.

Table 3
Cause of Death

Cause	BU-MAC (n = 14)	TBI-MAC (n = 45)
Leukemia	8 (57%)	23 (51%)
GVHD	1 (7%)	8 (18%)
Infections	1 (7%)	5 (11%)
Organ failure	1 (7%)	5 (11%)
Graft failure	2 (14%)	3 (7%)
Others	1 (7%)	1 (2%)

There are possible explanations for the differences between our findings and those of others. First, although previous studies showed that BU-MAC was better than TBI-MAC for AML patients in CR1 and for patients with myelodysplastic syndrome or chronic myeloid leukemia [11,12,14], there was no survival advantage for AML patients in CR2 [13]. Here, we undertook a combined analysis of AML patients in CR1/CR2, which may be a reason why i.v. BU-MAC failed to show superior survival rates. Also, the present study was underpowered with respect to detecting significant associations in the setting of CR1 alone. Second, the RFS rates in the TBI-MAC group in the present study were better than those reported by previous studies [13,14]. It is likely that the tight control of acute GVHD contributed to the low incidence of NRM in both the TBI-MAC and i.v. BU-MAC groups, resulting in favorable RFS rates. Because AML shows only modest sensitivity to acute GVHD and because limited sensitivity to chronic GVHD and relevant reductions in relapse rates were only observed in patients experiencing extensive chronic GVHD [27], the similar relapse rates and rates of extensive chronic GVHD observed in the 2 groups suggest that i.v. BU-MAC and TBI-MAC exert comparable antileukemic effects. Moreover, the lower NRM rates in the BU-MAC group may be due to factors such as younger age [13,28] and more patients being in CR1. The similar relapse rates between the groups may be because more patients were in CR1 but had unfavorable cytogenetics profiles in the BU-MAC group [29].

BU-MAC is believed to cause early toxicity and higher rates of sinusoidal obstruction syndrome than TBI-MAC [11]. However, Nagler et al. [26] showed that TBI- and BU-MAC carried a comparable risk of sinusoidal obstruction syndrome in AML patients. Intravenous BU-MAC appeared not to be a risk factor for transplantation when the i.v. dose was adjusted according to the target level of the drug or body weight. We found no

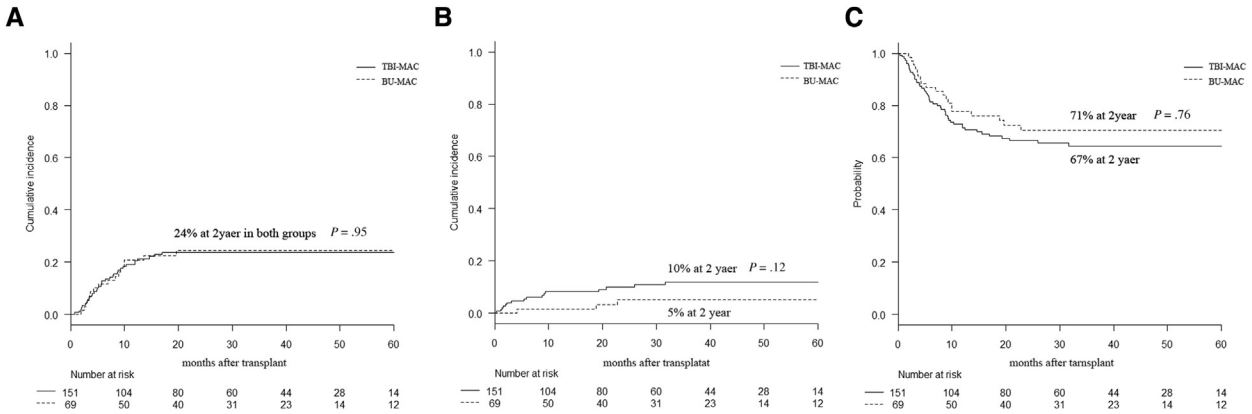

Figure 2. (A) The cumulative incidence of relapse. The 2-year cumulative incidence of relapse in the i.v. BU-MAC group (n = 69; dotted line) was 24% (95% CI, 15% to 35%) and in the TBI-MAC group (n = 151; solid line) was 24% (95% CI, 17% to 31%; $P = .95$). (B) Cumulative incidence of NRM. The 2-year cumulative incidences of NRM in the i.v. BU-MAC (dotted line) and TBI-MAC (solid line) groups were 5% (95% CI, 1% to 13%) and 10% (95% CI, 6% to 15%), respectively ($P = .12$). (C) Probability of RFS. The 2-year RFS rates in the BU-MAC (dotted line) and TBI-MAC (solid line) groups were 71% (95% CI, 58% to 80%) and 67% (95% CI, 58% to 74%), respectively ($P = .36$).

clear reason for the relatively low incidence of NRM in the i.v. BU-MAC group; indeed, both the incidence and cause of death were similar to those reported by Sisler et al. [13]. The follow-up duration was too short to allow us to compare the incidence of delayed complications between groups. However, previous reports suggest that BU-MAC is associated with lower rates of long-term morbidity such as growth and developmental problems, cataracts, hypothyroidism, hypertension, late cardiovascular events, and secondary malignant neoplasms [6-10]. These outcomes support the use of i.v. BU-MAC for pediatric patients, even if survival rates after i.v. BU-MAC are similar to those after TBI-MAC.

Here, we used CY in combination with TBI or MEL in combination with BU in most cases (77% and 73%, respectively) because HCT using a combination of BU plus MEL followed by an infusion of allogeneic marrow from an HLA-matched related donor achieves good RFS with minimal mortality and morbidity for patients with good-risk leukemia [30]. The AML-99 study (and the subsequent AML-05 study) in Japan recommended that patients in the CR1 setting receive a combination of BU plus MEL as a conditioning regimen if they had high- or intermediate-chromosome risk AML and an HLA-matched sibling donor [4]. This conditioning regimen has been widely used for AML patients in the CR1 setting in Japan. However, the present study (which comprised a small number of patients) failed to demonstrate that the combination of i.v. BU plus MEL was superior to i.v. BU plus CY, although neither showed excessive toxicity. On the other hand, a previous study reported that a combination of 3 alkylators (BU, CY, and MEL) was superior to other conditioning regimens (eg, BU and CY) for children with AML in the CR2 setting [31]. Furthermore, some studies report no significant difference in the relapse rate and a lower incidence of NRM or grade II to III regimen-related toxicity between BU-CY and fludarabine-BU regimens when used to treat patients with AML/myelodysplastic syndrome, especially in the CR1 setting [32,33]. Both CY and fludarabine are used primarily because of their immunosuppressive, rather than their antileukemic, properties (ie, to increase engraftment) [32]. Because MEL has profound stem cell toxic and immunosuppressive properties, we believe it is reasonable to replace CY with MEL plus BU for patients with malignancies whose origin is attributable to a hematopoietic stem cell defect (eg, AML) [31]. Further prospective and controlled trials are warranted to evaluate the optimal combinations of BU and other drugs for patients with AML.

In conclusion, the results of the present study suggest that i.v. BU-MAC is a promising alternative to TBI-MAC for pediatric AML patients in remission. Nonetheless, this retrospective registry-based analysis has several limitations. First, we do not know why individual patients were selected to receive specific conditioning regimens, and there were differences in the supportive care provided by individual physicians. Second, the lack of information about whether BU was targeted to a certain level may have skewed the results. This is because even when administered i.v., variable rates of BU clearance may result in either inadequate or excessive systemic exposure to the drug. Obtaining targeted BU area under the curve values via the i.v. route is fundamental to achieving therapeutic values and for preventing toxicity [34]. It appears likely that improvements in BU administration, including i.v. administration and dose adjustment, are responsible for the improved results observed with BU-MAC [12]. More recently, pediatric AML patients have received reduced-toxicity conditioning regimens. Antileukemic effects after allo-HCT conditioned by reduced-toxicity conditioning regimens are comparable with those after conditioning with standard MAC regimens [35,36]. Until a prospective trial is conducted, the observations reported herein support the use of i.v. BU-MAC rather than TBI-MAC for pediatric AML patients in remission.

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